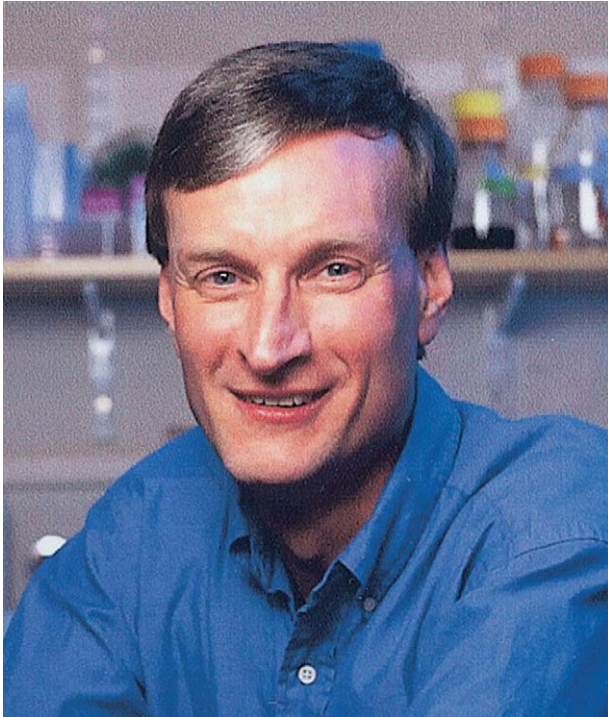


2003 CURT STERN AWARD ADDRESS On Low Expectations Exceeded; or, The Genomic Salvation of the Y Chromosome*

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David C. Page (courtesy Sam Ogden)

Receiving the Curt Stern Award makes me think of the year 1956, when Stern, an extraordinary *Drosophila* geneticist with broad-ranging interests, became president of the American Society of Human Genetics. It's also the year I was born. During the first year of my life, Stern presided over the Society's 10th annual meeting, in Ann Arbor. There, he delivered a presidential address entitled "On porcupine skin and hairy ears; or, the alleged sins

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of the Y chromosome." As usual, the editor of the *American Journal of Human Genetics* sanitized the title of Stern's address prior to publication ("The problem of complete Y-linkage in man" [Stern 1957]).

In his presidential address, Stern debunked all 17 "presumably or possibly Y-linked traits"—all flimsy claims based on shoddy pedigree analysis. By the end of his speech, no genes were left standing on the human Y chromosome. Even today, pedigree analysis has yet to reveal a single Y-linked gene. Surely, it wasn't Stern's intention, but his entirely accurate pronouncement of the futility of Y-linked pedigree studies led others to a new understanding: the Y chromosome must be a genetic wasteland. And so began the Rodney Dangerfield era for the human Y chromosome. Rodney has always felt a special kinship with this chromosome, frequently declaring, "Y don't get no respect!" In truth, Curt Stern was a fervent supporter and student of the human Y chromosome. He simply lacked the molecular and genomic tools required to understand it. So let's invite Stern back to enjoy this occasion.

First, for Stern's benefit, I would like to provide an executive summary of recent insights:

- Previously thought to be a genetic wasteland, the Y chromosome is now known to contain about 76 protein-coding genes. These genes collectively encode at least 27 distinct proteins. Many of these genes are functionally specialized in spermatogenesis (Lahn and Page 1997; Skaletsky et al. 2003).
- Today's X and Y chromosomes evolved from an ancestral pair of autosomes that existed 300 million years ago (Ohno 1967; Lahn and Page 1999).
- Once considered to be merely a rotting copy of that ancient autosome, the Y chromosome is now understood to have expanded its gene repertoire during evolution through (1) selective importation of genes from autosomes and the X chromosome and (2) gene amplification (Saxena et al. 1996; Skaletsky et al. 2003).
- Although researchers had believed the Y chromosome was composed almost entirely of junky repeats (even in its euchromatic portions), it actually contains gene-rich palindromes of unprecedented

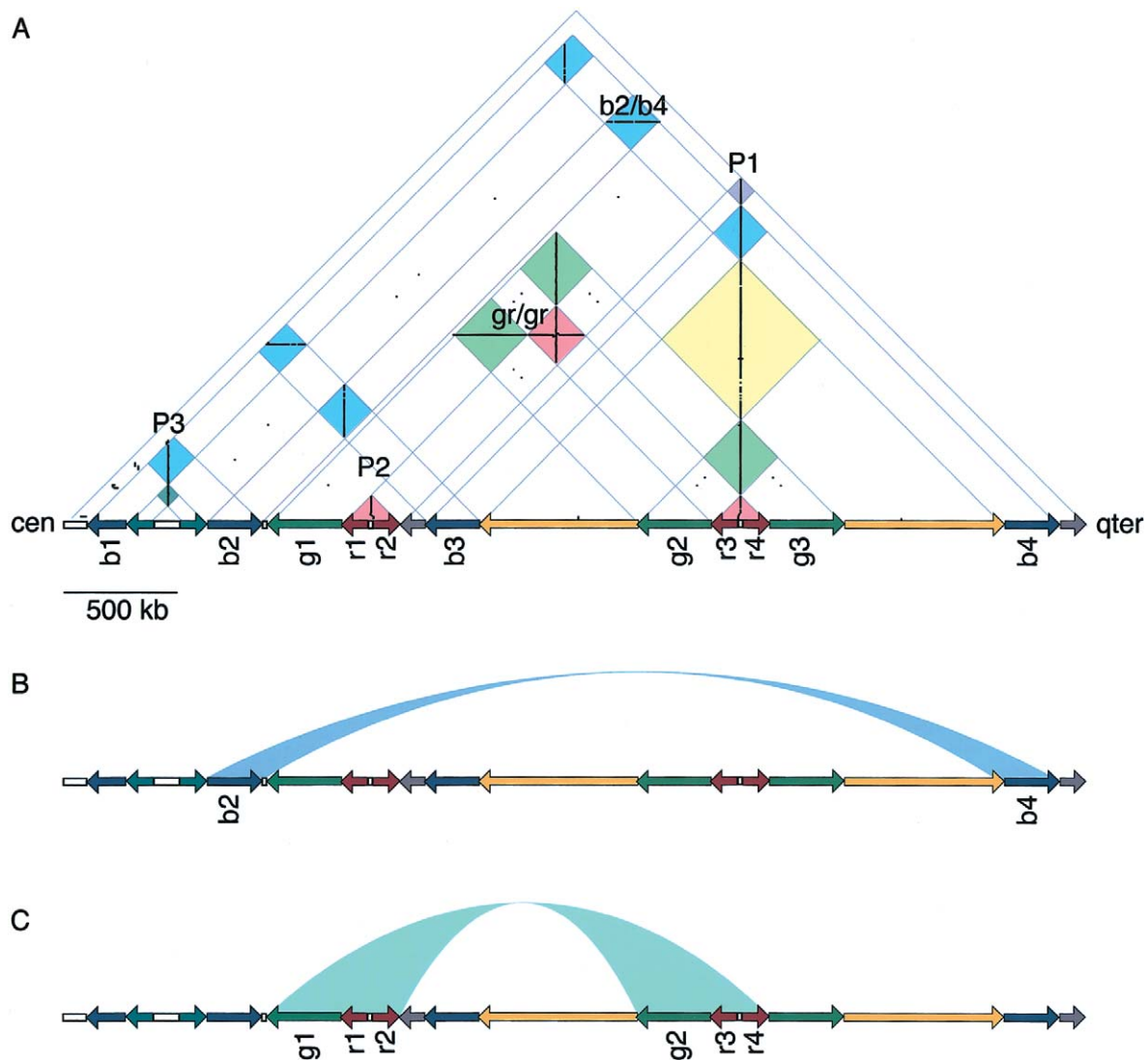


Figure 1 The Y chromosome's *AZFc* region, a deletion-prone hall of mirrors (adapted from Kuroda-Kawaguchi et al. [2001] and Repping et al. [2003]). *A*, Dot plot in which a 4.5-Mb portion of the human Y chromosome, including *AZFc*, is compared with itself. The base of the plot depicts the organization of amplified segments, or amplicons, including those labeled b1–b4 (blue), g1–g3 (green), and r1–r4 (red). Within the plot, each dot represents a perfect match of 500 bp. Direct repeats appear as horizontal lines, inverted repeats as vertical lines, and palindromes as vertical lines that nearly intersect the baseline. Palindromes P1, P2, and P3 are indicated, as are the direct repeats responsible for the b2/b4 and gr/gr deletions. Within the plot, tinted squares reflect pairs of amplicons with >99.9% identical sequences. Gray diagonal lines mark amplicon boundaries within the plot. Orientation with respect to centromere (cen) and long-arm telomere (qter) is shown. *B*, The blue arch highlights the regions demarcating the b2/b4 (*AZFc*) deletion. *C*, The green arch highlights the regions demarcating the gr/gr deletion.

scale and precision (Kuroda-Kawaguchi et al. 2001; Skaletsky et al. 2003).

- We no longer think of the Y chromosome as a land of no recombination and, hence, of inevitable gene decay. We now understand it to be a place of abundant gene conversion. Researchers have speculated that gene conversion helps preserve the integrity of Y-chromosome genes across evolutionary time (Rozen et al. 2003; Skaletsky et al. 2003).

Curt, let me show you the heart of the Y chromosome—the *AZFc* region—a spectacular hall of mirrors. The region is dominated by near-perfect sequence reflections, including three massive palindromes (the largest spanning nearly 3 Mb) as well as other inverted and direct repeats (fig. 1A). But in this reflected beauty lies great danger—the hall of mirrors is fragile. Particularly treacherous are the direct repeats, revealed by horizontal lines in the triangular dot plot of fig. 1A. One pair of

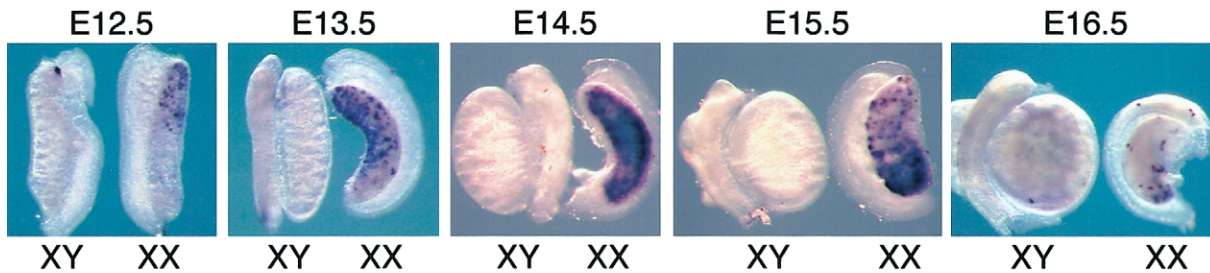


Figure 2 Expression of *Stra8* in XX and XY mouse embryonic gonads at 12.5–16.5 d following fertilization (from Menke et al. [2003], where details of this whole-mount in situ hybridization analysis can be found). Gonads are oriented with anterior poles at the top.

229-kb direct repeats (amplicons b2 and b4 in fig. 1B) form irresistible targets for homologous recombination, yielding de novo deletions of the intervening 3.5 Mb of DNA at a frequency of 1 in 4,000 male births (Kuroda-Kawaguchi et al. 2001).

Curt, as you speculated in your presidential address, the Y chromosome is loaded with fertility genes, and this is especially true in the *AZFc* region. Indeed, the recurrent b2/b4 (*AZFc*) deletion was the most common genetic cause of spermatogenic failure known—until this month, when Sjoerd Repping, Steve Rozen, and colleagues reported a far more common Y deletion, the *gr/gr* deletion (Repping et al. 2003).

The *gr/gr* deletion exists as a bona fide polymorphism in human populations, yet it tends to impair spermatogenesis. The *gr/gr* deletion removes half of the *AZFc* region (fig. 1C), reducing the copy number but not eliminating outright any of the region’s testis-specific gene families; two of four *DAZ* genes remain. The *gr/gr* deletion can be identified in ~1% of all men, making it perhaps 40 times as common as the b2/b4 (*AZFc*) deletion. Unlike the b2/b4 deletion, which almost always arises de novo, the *gr/gr* deletion is often inherited from the father. The *gr/gr* deletion increases the risk of spermatogenic failure, as demonstrated by association studies, but it does not routinely devastate spermatogenesis, as the b2/b4 (*AZFc*) deletion does. A true polymorphism, and a functionally important one at that, the *gr/gr* deletion represents the first meaningful convergence on the Y chromosome of medical genetics and population genetics (Repping et al. 2003). We may come to think of the *gr/gr* deletion as the Y chromosome’s $\Delta F508$, or APOE4.

Curt, in your presidential address, you also speculated that the human Y chromosome might have a role in sex determination, as was known in guppies and gypsy moths. You were right! In the developing human embryo, the gonad is the first structure to differentiate, anatomically, between XX and XY. Through work that began with Charles Ford (Ford et al. 1959) and Patricia Jacobs (Jacobs et al. 1959) and culminated in the laboratories of Peter Goodfellow and Robin Lovell-Badge, we now know

that a single gene on the Y chromosome—*SRY*—instructs the gonads to develop as testes (Sinclair et al. 1990; Koopman et al. 1991). Built around this knowledge is much additional genetic insight into the making of testes—and males. But here’s the rub. Embarrassingly, in the year 2003, we know little genetically about the making of the ovary, about how an XX embryo begins to become a phenotypic female. We shamelessly legitimize our ignorance by referring to the female pathway as the “default.” And so I pose a question for the future: what is the role of genes in the making of the ovary?

I would like to offer an early observation that pertains to this question. Curt, we now think of genes not only as sites of heritable variation underlying phenotypes but also, through their RNA or protein products, as markers of cellular identity during development. My colleague Douglas Menke identified a gene, *Stra8*, whose expression pattern in embryonic gonads is intriguing in this light. Shown in figure 2 is the localization of *Stra8* RNA in mouse gonads at 12.5–16.5 d after fertilization. *Stra8* is expressed in XX (but not XY) embryonic gonads, in a slowly advancing wave that begins at ~E12.5, near the anterior pole of the embryonic ovary, and ends at ~E16.5, near the posterior pole. Other experiments have shown that *Stra8* is expressed only in germ cells, not in the somatic cells of the gonad. At present, *Stra8* expression in germ cells is the earliest known marker of female differentiation in mammals (Menke et al. 2003). These findings raise the possibility that the first steps in turning an XX embryo into a phenotypic female may unfold in germ cells. Much work remains to be done!

It is a delight to know that this vibrant community of scholars thinks well enough of my laboratory’s work to honor me with the Curt Stern Award. At its best, science reflects not an individual effort, but a collective one, with a sense of mission and purpose shared among colleagues and shared between mentors and students. I am fortunate to have worked with an exceptional group of mentors, colleagues, and students throughout my career. I would like to express my appreciation to these men and women, too numerous to mention, whose creativity and

perseverance are honored here today. Finally, and most especially, I'd like to remember the source of my own Y chromosome, a man who has been my greatest mentor and my greatest appreciator: Conrad Page, Jr. (September 20, 1920–October 17, 2003). The 2003 Curt Stern award is for you, Dad.

Acknowledgments

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